

Antinociceptive And Antipyretic Properties Of *Tapinanthus Dodoneifolius* Leaf Extract In Albino Rats

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Abstract

The present study was designed to investigate the analgesic and antipyretic effects of the ethanolic extract of dried leaves of *Tapinanthus dodoneifolius* (DC, VANTIEGH) in rats using three laboratory models: brewers' yeast induced hyperpyrexia test, formalin-induced paw licking and hot plate latency test respectively. The extract (25-200mg/kg) caused a significant ($P < 0.05$) decrease in body temperature of the animals. The extract (25-200mg/kg) also significantly ($P < 0.05$) increased the reaction time of the animals to thermal stimuli and also significantly ($P < 0.05$) decreased both the early and late phases of formalin induced paw licking in rats. This indicates that the extract increased pain threshold in the hot plate latency test and in the early and late phases of the formalin-induced paw licking in rats. The result also indicated an ability of the plant to reduce pyrexia in albino rats.

Keywords: *Tapinanthus dodoneifolius*, Antipyretic, Pain threshold, Rats

Introduction

Tapinanthus dodoneifolius (DC, Vantiegh), is a parasitic plant of West-African origin. It is one of the medicinal plants used locally for the treatment of several human and animal ailments¹. The leaves and young twigs of the plants have also been used in folklore medicine to treat different diseases such as circulatory and respiratory diseases, malaria, diabetes, hypertension and sterility². The cardiovascular, spasmolytic and anti inflammatory activities of water extract of *A. dodoneifolius* have been reported³. Cepleanu *et al.*⁴ also reported the larvicidal and molluscicidal activities of this plant. Preparations of *Tapinanthus dodoneifolius* have been used in the traditional Nigerian medicine to treat malaria and this practice has remained till date without scientific validation⁵.

Pain has been defined by IASP⁶ as an unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage. Pain, inflammation and fever usually have some molecular mechanism in common especially through the release prostaglandins. This form the basis for the use on non steroidal anti-inflammatory drugs (NSAIDs)⁷. Furthermore, pain usually accompanies fever and treatment of pain symptoms with analgesics may provide temporary relief for individuals suffering from fever⁸. Therefore, it might be either the pain or fever component that the extract of *Tapinanthus dodoneifolius* ameliorates when feverish individuals administer the extract therapeutically. However, there is a dearth of information on the effect of this plant extract on pain threshold and pyrexia. Therefore, the present study was carried out to investigate the anti-nociceptive and antipyretic properties of the leaf extract of the plant in rats in accordance with its ethnomedicinal uses by local folks.

Materials And Methods

Animals

Male and female Wistar rats weighing 200-230 g were used for the study. The animals were bred and housed in the animal house of the Faculty of Basic Medical Sciences, University of Ilorin. The animals were kept in clean, standard cages that are well ventilated. They were provided with mouse cubes (Bendel feeds) and water *ad libitum* prior to the commencement of the antipyretic and analgesic studies.

The research was conducted in accordance with the ethical rules on animal experimentation, and approved by the use of animal ethical committee of the University of Ilorin.

Plant and extract

Tapinanthus dodoneifolius leaves were collected in Tanke area, Ilorin, Kwara State, Nigeria. The plant was identified by Mr Felix Inah of the Forestry Research Institute of Nigeria (FRIN, Ibadan) where a voucher specimen with herbarium file number FHI 107663 was deposited.

The leaves of *T. dodoneifolius* were air-dried, reduced to powdered form and sieved to obtain fine powder.

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500g of the powder was put in and soaked in 2,500ml ethanol, shaken and allowed to stand for 24hours. The macerated mixture was filtered and evaporated in a carefully regulated water bath maintained at temperature of 50°C to yield a deep green semi-solid extract weighing 39.5g which was stored at 4°C in a refrigerator.

Experimental Procedure

Antipyrexia study

Brewers' yeast induced hyperpyrexia test

This study was carried out by slightly modifying the method described and employed by Sharma et al.¹. The animals were divided into six groups A-F (comprising five animals each). Pyrexia was induced by subcutaneously injecting 20% (w/v) brewers' yeast suspension (10ml/kg) into the dorsum of the rats. 17hours after injection, the rectal temperature was measured using clinical digital thermometer. Rats that showed increase in temperature of at least 0.7°C were used for the test. Rats grouped A-D were given 25, 50, 100 and 200 mg/kg of extract orally, which represent very low, low, medium and high dose respectively. Rats in group E and F were given normal saline (10ml/kg) and indomethacin (5mg/kg) respectively. The initial rectal temperature of the rats prior to brewers' yeast injection was measured and compared with the temperature measured at 60, 90, and 120 minutes after each administration of the respective oral solutions.

Antinociceptive study

Hot plate latency test

This was carried out by slightly modifying the method used previously¹⁸. The rats were placed on hot plate maintained at 55°C ± 1 °C and confined with a carbon box 50cm high. The time taken for the rat to jump off the plate was recorded as the latency in seconds. The

rats used for this study were fasted 12 hours before the test and subsequently divided into six groups (A-F). Rats in B, C, D, and E were given 25, 50, 100 and 200 extract of *T. dodoneifolius* orally after twelve hours fast respectively. Rats in group A and F were given doses of normal saline (10mg/kg) and indomethacin (5mg/kg) respectively. One hour after extract/drug administration, each rat was placed on the hot plate and the latency time recorded, subsequent measurements were conducted at 30, 60 and 90 minutes respectively for all groups.

Formalin induced paw licking test

This was carried out by method previously described¹⁹. The rats were divided into six groups (A-F). Rats in B, C, D, and E were given 25, 50, 100 and 200 extract of *T. dodoneifolius* orally after twelve hours fast respectively. Rats in group A and F were given doses of normal saline (10mg/kg) and indomethacin (5mg/kg) respectively. One hour after extract/drug administration, formalin was injected into the left hind paw, of the animals and the time spent by each rat in licking the injected paw in the early (0-5 minutes) and late (20-30 minutes) phases were recorded.

Statistical analysis

In these studies values are expressed as mean ± standard error of mean (SEM). Statistical significance was determined using the ANOVA followed by Duncan post hoc test and values were accepted as significant when $p < 0.05$.

Results

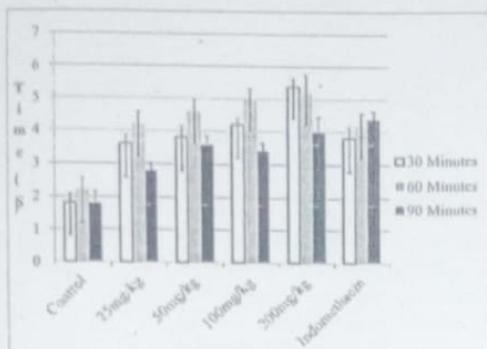
Antipyrexia study

The 25 mg/kg body weight of the ethanolic extract of *T. dodoneifolius* leaves significantly reduced the elevated rectal temperature from 37.2±0.12 to 36.8±0.01 after 90 minutes of its administration. All the

Table 1: Effect of ethanolic extract of *T. dodoneifolius* on brewers' yeast induced hyperpyrexia test.

Group	Dose (mg/kg) Orally	Temp.(°C) after 17 hours of yeast injection	Temp.(°C) at different time intervals after saline/extract administration		
			60mins	90mins	120mins
Control (Normal saline)	10	37.75±0.1	37.62±0.11	37.41±0.16	37.32±0.18
<i>T. dodoneifolius</i>	25	37.20±0.12	37.0±0.13	36.80±0.01 *	36.60±0.09 *
<i>T. dodoneifolius</i>	50	37.57±0.17	37.17±0.20	36.76±0.227 *	36.40±0.31 *
<i>T. dodoneifolius</i>	100	38.04±0.127	37.44±0.225	37.05±0.29 *	36.83±0.38 *
<i>T. dodoneifolius</i>	200	38.20±0.21	37.6±0.14 *	37.30±0.01 *	37.0±0.16 *
Indomethacin	5	36.0±0.15	36.21±0.38 *	35.20±0.39	34.80±0.40

Each value represents the mean ± SEM for 5 rats. *P < 0.05 compared to temperature 17 hours after yeast injection



higher doses of the extract and indomethacin follow the same pattern as shown in table 1

Antinociceptive study

Hot plate latency test

The results of this study is shown in figure 1 and it shows that the extract significantly increase the reaction time of the animals to thermal stimuli starting from 30 minutes to 60 minutes compared to the control. The highest dose (200 mg/kg body weight) produced consistent anti nociception on the hotplate compared with the control and other groups.

Formalin induced paw licking test

In figure 2, the 25 mg/kg ethanolic extract of *T. dodoneifolius* significantly ($p < 0.05$) reduced the time spent on paw licking from 131.2 ± 13.6 to 93.2 ± 6.1 and 159.8 ± 7.1 to 105.8 ± 7.7 in the early and late phases of the test respectively. The higher doses followed the same pattern.

Discussion and Conclusion

The present study investigated the effects of *T. dodoneifolius* on pain threshold and pyrexia using common animal models. The results showed that *T. dodoneifolius* extract has significantly increased pain threshold using the hotplate as well as the formalin models. This indicates that the extract can inhibit pain resulting from direct neuronal stimulation (formalin, first phase) and those arising from inflammatory processes (Formalin, second phase)¹¹⁻¹³. Result from the brewers yeast also show that the extract could significantly reduce experimentally induced pyrexia although the mechanism is not clearly understood.

It is not surprising that *T. dodoneifolius* has antinociceptive effects especially a direct effect on the nervous system as represented by the activity on the thermal test and first phase of the formalin test^{11,13} because previous reports show that the plant has the ability to calm nerves and heart problems among other properties^{12, 14-15}. Indeed, the thermally induced pain has been designated as a form of central pain involving

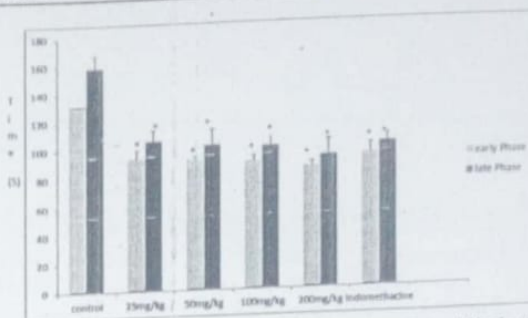


Figure 2. Effect of *T. dodoneifolius* extract on formalin induced paw licking in rats. * $P < 0.05$ compared with control group

modulation of pain pathways not only in the peripheral areas but in spinal cord and agents/drugs that have ability to reduce the reaction time in this model have the potential to be strong analgesics¹⁶⁻¹⁷.

The brewer's yeast is a common and inexpensive model for assessing antipyretic activities in rodents. The weakened yeast is a source of external pyrogen which has the capacity to trigger the endopyrogens and ultimately stimulate measurable pyrexia by increasing prostaglandins E_2 . In the present study, it was observed that the extract even at the lowest dose was able to significantly reduce the pyretic state of the animals although after 9 minutes of administration. This shows that the extract at low dose is effective but it has the slightly delayed onset of action compared with the highest dose which is effective even from 30 minute after its administration to pyretic animals.

Generally the trend of the results indicate that there might be a common mechanism for the activities of this plant extract. This mechanism was not examined in the present study, however, it might not be unconnected with inhibition of cyclooxygenase (COX) which the NSAIDs inhibits to achieve their therapeutic effects. NSAIDs have moderate analgesic, anti-inflammatory and antipyretic effects linked to inhibition of COX and ultimately prostaglandins synthesis⁷.

Furthermore, medicinal properties of plants and natural substances are usually derived from their constituents especially the secondary metabolites and this would be the case for *T. dodoneifolius*. Fasanu and Oyedepo¹⁸ reported that the plant contains phytochemicals like alkaloids and saponins. The results of the mineral analysis of the major cations also show that the leaves, the stem bark of the host plants and the mistletoe growing on them contain major cations like potassium, magnesium, calcium and sodium in varying quantities. Therefore, the probable mechanism underlying the activities of the plant might

be associated with its active agents. Previous studies have shown the active agents to be alkaloids, saponins and tannins ". Phytochemicals help plants defend against environmental challenges and also provide humans with protection against various diseases as well. Hence it is not surprising while they are harvested for medicinal purposes. These compounds have the ability to reduce pain perception in the models used.

In conclusion, this study has shown that ethanol extract of *T. dodoneifolius* has the capacity to increase pain threshold and reduce pyresis in animals which might be based on its phytochemical constituents. Further studies are required to isolate and characterise the active ingredients in the plant.

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